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The Effect of Rapid Chilling of Pork Carcasses during the Early Postmortem Period on Fresh Pork Quality

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Abstract: The objective was to investigate the effects of blast chilling on pork quality in cuts from the *Longissimus thoracic et lumborum* (LM), *Psoas major* (PM), *Semimembranosus* (SM; both superficial [SMS] and deep [SMD] portions) and the *Triceps brachii* (TB). Forty carcasses (10 carcasses per replication) were split and sides were assigned to either blast chill (BC, –32°C for 90 min, followed by spray chill at 2°C for 22.5 h) or conventional chill (CC, spray chilled at 2°C for 24 h) regimens. The LM from BC sides had lower ($P < 0.05$) temperature at 2 h postmortem (CC 21.8°C, BC 9.7°C), 4 h (CC 13.3°C, 3.8°C BC), 22 h (CC 4.2°C, BC 1.4°C), and 30 h (CC 0.4°C, BC –0.2°C). The LM pH in BC sides was higher at 4 h (CC 6.09, BC 6.34), 22 h (CC 5.81, BC 5.89), and 30 h (CC 5.68, BC 5.74) postmortem. The BC resulted in higher ($P < 0.05$) 30 h postmortem pH in the SM compared to the CC regime (CC 5.68, BC 5.74). The BC sides had increased ($P < 0.05$) purge in the PM (CC 0.48%, BC 0.74%) and increased ($P < 0.05$) cook loss in chops from the LM (CC 22.37%, BC 24.24%). The PM from BC sides were more juicy (CC 7.50, BC 8.30), less chewy (CC 2.80, BC 2.10), and more tender (CC 7.90, BC 8.60). Chops from the LM of BC sides had greater Warner-Bratzler shear force (CC 2.00, BC 2.30). Color was affected in the SM with BC sides showing darker color score (CC 3.00, BC 3.20) and redder Hunter a value (CC 16.35, BC 16.02). Chilling treatment did not affect sarcomere length in the LM. Treatment did not affect postmortem proteolysis in any cut. The response to chilling regimen is different across different muscles which may be caused by location, rate of chilling, and fiber type.

Keywords: pork chilling, pork quality, protein degradation, tenderness
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Introduction

Leaders in the meat industry now realize that importance of meeting consumer demands for tender, palatable, and safe fresh pork products as emphasis shifts from a commodity to a product focus in the global marketplace. Therefore, all factors that influence these characteristics must be defined. Once the slaughter process begins, one of the first impacts on pork quality is the reduction of carcass temperature. This step in the process is vital to reduce microbial growth. The rate and extent of carcass chilling has been shown to negatively impact fresh pork quality by influencing sarcomere length and postmortem proteolysis (Bendall, 1975; Shackelford et al., 2012). Color and water holding capacity are improved with the implementation of rapid chilling systems (Jones et al., 1993; Janiszewski et al., 2018). Rybarczyk et al. (2015) reported loins of blast chilled carcasses had less drip loss, but were less tender compared with conventional chilling. Rosenvold et al. (2010) showed that a slower, stepwise chilling method improved tenderness in loin chops compared with chops from conventionally chilled carcasses. However, Rosenvold et al. (2010) did not report a difference in thaw loss or cook loss in response to stepwise chilling method. Differential response in tenderness and water holding capacity have also been reported in the ham (Ohene-Adjei et al., 2002; Springer et al., 2003).

Rate of chilling is also affected by carcass size (Overholt et al., 2019) and therefore carcass size is also a source of variation in chilling even when the chilling system is constant. Due to location and fiber type differences across muscles, the effect of chilling will not have a uniform impact on fresh pork quality. The effect of chilling method across different muscles (*Longissimus thoracic et lumborum, Semimembranosus,* and *Biceps femoris*) has been determined (Rosenvold et al., 2010), but it is not yet well defined. Therefore, the objectives of the current study were to (1) define the effect of blast chilling on fresh pork quality in cuts from the *Longissimus thoracic et lumborum, Psoas Major, Semimembranosus,* and the *Triceps brachii* and (2) characterize the effect of chilling on muscle location in the *Semimembranosus* superficial and deep portions.

Materials and Methods

Carcasses were selected in a commercial facility under the supervision of the USDA Food Safety Inspection Service. The experiments described here were conducted on the carcasses and therefore no animal care protocol was necessary. Pork carcasses (n = 40) were selected in groups of 10 on 4 nonconsecutive slaughter dates. Carcasses were selected on the processing floor, immediately following evisceration. Criteria for selection included hot carcass weight (HCW; 86 to 91 kg) and fat-free lean (FFL; 54 to 57%). After selection, carcasses were split and alternating sides assigned to blast chill (BC) or conventional chill (CC) treatment approximately 45 min postmortem. Sides that received BC treatment were subjected to –32°C chilling cycle for 90 min followed by spray chilling and storage at 2°C. Sides assigned to CC were spray chilled and held at 2°C. Loin temperature was measured and continuously recorded at the region of the 10th rib on 2 sides from each treatment at each slaughter date using LogTag data loggers (LogTag Recorders Limited, Auckland New Zealand). Tenth rib LM pH was recorded in each carcass entering chilling treatment, approximately 4, 22, and 30 h postmortem. Tenth rib loin temperature measurements were recorded entering chilling treatment, exiting chilling treatment (for BC sides) and approximately 135 min postmortem (CC sides), and at 4, 22, and 30 h postmortem. All carcasses were held at 2°C overnight and processed the following day.

Sample collection

The *Longissimus thoracic et lumborum* (LM), *Triceps brachii* (TB), *Semimembranosus* (SM) and *Psoas major* (PM) were collected from each carcass approximately 26 h postmortem. All cuts were vacuum packed and transported on ice to the Iowa State University sensory evaluation lab. Fresh pork was aged (PM 7 d, SM 8 d, LM 10 d, and TB 13 d) under refrigeration (4°C) and used to determine treatment effect on ultimate pH, purge loss, cook loss, star probe, Warner-Bratzler shear force (WBS), and sensory quality. Purge measurements were collected following aging. Samples were removed from their vacuum bag and weighed. The weight of the bag was then subtracted from the weight of purge remaining in the bag and purge was determined as a percentage of the original muscle weight [(initial weight – final weight)/initial weight] × 100. Ultimate pH was recorded on whole cuts and 2.54 cm thick chops (LM and SM) were prepared. Hunter L, a, and b values were collected using a Minolta Chroma Meter fitted with a 50 mm aperture and a D65 light source with a 2° observer. Color scores (National Pork Board, 1999; standard 6-point scale, 1 = pale pinkish gray to white; 6 = dark purplish red) marbling scores (National Pork Board, 1999; standard 10-point scale, 1 = 1.0% intramuscular fat; 10 = 10.0% intramuscular fat) were assigned to chops (SM and LM).

Sensory analysis

Raw weights were collected on chops (LM and SM) and roasts (PM and TB) for cook loss determinations. Chops were cooked in clamshell grills to an internal temperature of 68°C (Santos et al., 2018). Roasts (PM and TB) were cooked in a rotary hearth oven at 176°C to an internal temperature of 68°C. Cooked weight was then collected and samples were divided into 1.27 × 1.27 × 2.54 cm cubes for trained sensory analysis (n = 4; Carlson et al., 2017a). Two additional chops or roasts were cooked using the same procedures as listed above. These chops were allowed to cool to 22°C. An Instron Universal Testing Machine (Instron Industrial Products, Grove City, PA) fitted with Warner-Bratzler shear force head was used to determine Warner-Bratzler shear force values (Schulte et al., 2019). A 5-pointed star probe was fitted to the Instron and compressed the sample to 80% of its height to determine star probe value (Lonergan et al., 2007; Schulte et al., 2019).

Sensory analysis was conducted as described by Carlson et al. (2017a) to determine tenderness, chewiness, juiciness, flavor, and off-flavor. Trained panelists

($n = 4$) evaluated samples on a 10-point anchored scale. A lesser value represents a lesser degree of a trait and greater values represents a greater degree of the trait (1 = tough, 10 = tender; 1 = not chewy, 10 = chewy; 1 = dry, 10 = juicy; 1 = no pork flavor, 10 = intense pork flavor; 1 = no off-flavor, 10 = intense off-flavor). Panelists were required to complete a 6-mo training period. During this period, they were provided with samples that exhibited extremes of the sensory attributes measured in each cut included in the study. Panelists were served 1.27 × 1.27 × 2.54 cm cubes of each sample immediately following cooking. Unsalted crackers and ice water were provided between samples.

Whole muscle protein extracts

Samples aged 2 and 10 d postmortem were frozen in liquid nitrogen and then powdered using a Waring blender (Waring Commercial, New Hartford CT). Whole muscle protein was extracted from frozen powdered samples as described by Carlson et al. (2017a, 2017b) using a solubilizing buffer (2% wt/vol SDS, 10m*M* sodium phosphate, pH 7.0). Protein concentration was determined using a Lowry protein assay with premixed reagents (DC protein assay, Bio-Rad Laboratories, Hercules, CA). Protein solutions were adjusted to 4 mg/mL in Wang's tracking dye (3 mM EDTA, 3% [wt/vol] SDS, 30% [vol/vol] glycerol, 0.01% [wt/vol] pyronin-Y, 30 mM Tris HCl, pH 8.0), vortexed, heated (approximately 50°C for 15 min), and then stored at –80°C.

SDS-PAGE and Western Blot analysis

SDS-PAGE and Western Blot were all conducted as described by Carlson et al. (2017a, 2017b). Briefly, desmin degradation on all samples (PM, TB, SMS, SMD, and LM) aged 2 and 10 d postmortem was determined using 10% polyacrylamide separating gels (acrylamide/bisacrylamide = 100:1 [wt/wt], 0.1% [wt/vol] SDS, 0.05% [vol/vol] tetramethylenediamine [TEMED], 0.05% [wt/vol] ammonium persulfate [AMPER], and 0.5 M Tris HCl pH 8.8). Troponin-T degradation was determined on LM samples aged 2 and 10 d using 15% polyacrylamide gels. Autolysis of calpain-1 in LM samples aged 2 d was determined using 8% polyacrylamide. For desmin degradation and calpain-1 autolysis analysis, 40 µg of sample was loaded in each lane, while 20 µg of sample was used in troponin-T degradation determination. Gels (10 cm wide × 8 cm tall) were run in SE 260 Hoefer Mighty Small II (Hoefer, Inc., Holliston, MA) electrophoresis units. The running buffer was a solution composed of 25 m*M* Tris, 192 m*M* glycine, 2 m*M* EDTA, and 0.1% [wt/vol] SDS. Desmin gels were run for approximately 375 V-hours, troponin-T gels were run for approximately 360 V-hours and µ-calpain gels were run for approximately 350 V-hours. Proteins from gels were transferred to polyvinylidene difluoride (PVDF) membrane. Blots were blocked for 1 h at 22°C using 5% non-fat dry milk [wt/vol] in PBS-Tween (80 m*M* Na_2HPO_4, anhydrous, 20 m*M* NaH_2PO_4, 100 m*M* NaCl, 0.1% [vol/vol] polyoxyethylene sorbitan monolaurate [Tween-20]. Blocking solution was then discarded and primary antibodies were added to PBS-Tween in the following dilutions, desmin- 1:40,000 using rabbit anti-desmin polyclonal antibody produced in house, troponin-T- 1:40,000 using monoclonal mouse anti-troponin-T (T6277, JLT-12, Sigma, Saint Louis, MO) and µ-calpain- 1:5,000 using monoclonal mouse anti-µ-calpain (MA3–940, Thermo Scientific, Rockford, IL). Blots were incubated in 10 mL of diluted antibody solution overnight at 4°C. Blots were then washed 3 times in 10 min intervals using 10 mL of PBS-Tween before a 1 h incubation in secondary antibody (22°C). Dilutions for secondary antibodies in PBS-Tween are as follows: desmin- 1:20,000 using goat anti-rabbit antibody (31460, Thermo Scientific), troponin-T- 1:30,000 using goat anti-mouse antibody (A2554, Sigma), and µ-calpain- 1:10,000 using goat anti-mouse antibody (A2554, Sigma). Following secondary antibody incubation, blots were again washed 3 times in 10 min intervals using 10 ml of PBS-Tween before detection using a chemiluminescent detection kit (ECL Prime, GE Healthcare, Piscataway, NJ).

Densities of immunoreactive protein bands were quantified using ChemiImager 5500 (Alpha Innotech, San Leandro, CA) and Alpha EaseFC (v. 2.03; Alpha Innotech). Desmin degradation was indicated by a decrease in the intensity of the approximate 55 kDa band and compared against a reference collected from a pork LM aged 7 d postmortem, troponin-T degradation was indicated by an increase in intensity of the approximate 30 kDa band and compared against a reference collected from a pork LM aged 7 d postmortem, and 2 d µ-calpain autolysis was indicated by an increase in the approximate 76 kDa band and compared to LM reference aged 12 h postmortem.

Calpastatin assay

Calpastatin activity was determined on LM samples at 2 d postmortem following procedures of Cruzen et al. (2014). Fresh muscle was finely minced and 10 g was homogenized in 30ml extraction buffer (100 m*M* Tris-HCl pH 8.3, 10 m*M* mercaptoethanol,

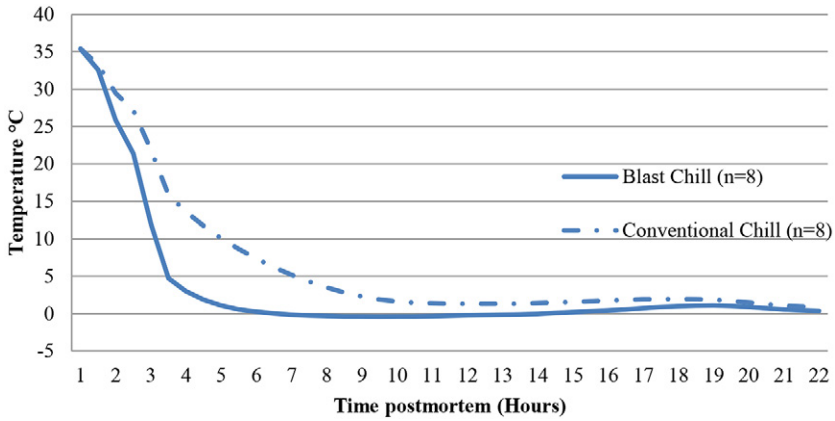

Figure 1. Temperature decline of *Longissimus dorsi* (LM) as measured at the tenth rib with continuous measurements. * Means represent 8 sides from each treatment group. Mean temperatures collected on 4 non-consecutive slaughter dates with 2 sides from each treatment per slaughter.

10m*M* EDTA, 100mg/L ovomucoid) using a Polytron PT 3100 (Lucerne, Switzerland). The solution was then centrifuged at 25,000 × *g* for 20 min and filtered through cheesecloth. Samples were then dialyzed for at least 6 h against 40ml of 40m*M* Tris-HCl pH 7.4, 1m*M* EDTA. Following dialysis, samples were heated for 20 min in a 95°C water bath and immediately chilled in ice cold water for 15 min. Samples including proteins in suspension and solution were transferred to 30 mL centrifuge tubes, centrifuged at 25,000 × *g* for 20 min, filtered through cheesecloth and volumes recorded for calpastatin activity calculation. Calpastatin activity was using partially purified lung m-calpain (Koohmaraie, 1990; de Oliveira et al., 2019).

Sarcomere length determination

One LM chop was collected 30 h postmortem, frozen and stored until analysis. Powdered frozen muscle tissue was suspended in a sucrose buffer (20 mM Sucrose, 20 mM potassium chloride, 5 mM sodium iodoacetate; pH 7.0) on the slide. Approximately 50 ML of the homogenate was placed on a slide with a cover slip. The beam of a helium-neon laser (γ = 0.6328 nm; Melles Griot, Carlsbad, CA) was passed through the homogenate and the distance between first-order diffraction bands was recorded. Sarcomere length was calculated using the equation prescribed by Cross et al. (1981). Five replicates were conducted for each sample.

Statistical analysis

Data were analyzed using a mixed linear model (PROC MIXED, SAS Enterprise Guide 5.1, SAS Inst. Inc., Cary, NC). The model included fixed effects of Treatment (TRT), Harvest day, and TRT*Harvest day. Carcasses (n = 40) served as random effects. To determine the effect of carcass composition, covariates of HCW, FFL, and the quadratic of each were added to the model. Least square means and standard errors were reported for each trait. A model using fixed effects of TRT, Harvest day, TRT*Harvest day, Location (SMS vs. SMD), and Location*TRT and carcass as a random effect was used to determine the effect of location within the SM. Significance level was determined at $P \leq 0.05$.

Results and Discussion

Average HCW was 89.0 ± 1.3 kg, average fat free lean (FFL) was 56.4 ± 0.8% across carcasses (n = 40). Continuous measurements of temperature decline in the LM at the tenth rib are represented in Fig. 1. Additional LM temperature measurements were taken on entering chilling treatment, exiting chilling treatment, approximately 4, 22, and 30 h postmortem and are represented in Table 1. Temperature decline of the LM in BC sides was more rapid than CC sides. Sides had similar temperature when entering the chilling environment. However, BC sides had lower temperature exiting chilling treatment (approximately 2 h postmortem in CC sides) 4, 22, and 30 h postmortem temperature. Results of BC in this experiment showed similar temperature declines reported by Shackelford et al. (2012) who showed a 4 h loin temperature of approximately 2.5°C. Rybarczyk et al. (2015) used blast chilling, but did not show similar rates of temperature decline with a 6 h loin temperature of 8.27°C.

The LM of BC sides had higher pH at 4, 22, and 30 h postmortem (Table 1). No significant treatment effect

Table 1. Effect of chilling on temperature and pH decline in the *Longissimus thoracic et lumborum*[1]

Time postmortem	Conventional chilled sides	Blast chilled sides	SEM	*P*-value
	pH			
Entering chilling treatment	6.57	6.52	0.22	0.44
4 h	6.09	6.34	0.07	< 0.0001
22 h	5.81	5.89	0.02	0.01
30 h	5.68	5.74	0.02	0.02
Ultimate[2]	5.68	5.68	0.03	0.86
	Temperature			
Entering chilling treatment	37.9°C	38.0°C	0.97	0.75
Exiting chilling treatment[3]	21.8°C	9.7°C	2.45	< 0.0001
4 h	13.3°C	3.8°C	1.48	< 0.0001
22 h	4.2°C	1.4°C	1.15	< 0.0001
30 h	0.4°C	–0.2°C	0.07	< 0.0001

[1]Means reported from LM of BC (n = 40) or CC (n = 40) sides.

[2]Ultimate pH represent pH measurement taken 10 d postmortem.

[3]Approximatley 135 min postmortem.

Table 2. Effect of chilling on pH in cuts from the *Psoas major, Triceps brachii,* and *Semimembranosus*[1]

Muscle	Conventional chilled sides	Blast chilled sides	SEM	*P*-value
30 h postmortem pH				
Psoas major	6.00	6.05	0.05	0.29
Triceps brachii	5.99	5.96	0.05	0.56
Semimembranosus	5.68	5.73	0.02	0.002
Semimembranosus superficial	5.70	5.74	0.02	0.11
Semimembranosus deep	5.66	5.72	0.02	0.01
	(0.05)[3]	(0.43)[3]		
Ultimate pH[2]				
Psoas major	6.01	6.02	0.05	0.90
Triceps brachii	6.01	6.00	0.05	0.86
Semimembranosus	5.77	5.80	0.04	0.47

[1]Means reported of pH from BC (n = 40) or CC (n = 40) sides.

[2]Ultimate pH represents pH on day of sensory analysis (PM 7 d, SM 8 d, TB 13 d).

[3]*P*-value representative between muscle locations. Fixed effects of TRT, harvest, harvest*TRT, muscle, and muscle*TRT with carcass serving as random effect used for analysis.

was observed in ultimate (10 d) pH in chops from the LM. Ultimate pH was not affected by chilling rate in previous studies (Springer et al., 2003; Shackelford et al., 2012; Rybarczyk et al., 2015). The results from the current experiment confirm those results and suggest that rate of pH decline is influenced by rapid chilling, a hypothesis supported by Kylä-Puhju et al. (2005).

Blast chilling resulted in higher pH in the SM at 30 h postmortem (Table 2). Chilling had no effect on ultimate (8 d) pH in the SM. Previous reports (Jones et al., 1993; Rosenvold et al., 2003; Tomović et al., 2008) also demonstrated that similar blast chilling regimens had no effect on 1 d postmortem SM pH when compared with slower chilling methods. In the current study, the SM was divided into a superficial (SMS) and a deep (SMD) portion to determine the effect of chilling on different locations throughout the muscle. Chilling regimen did not have an effect on 30 h pH in the SMS or SMD. Analysis of 30 h pH in SMS and SMD did however find a significant difference between the SMS and SMD of CC sides while this difference was not found in BC sides. It is likely that the superficial portion chilled at a more rapid rate than the deep portion due to more direct exposure to air temperature resulting in decreased pH decline. This result may also demonstrate the insulating effect of the outer portion of the muscle.

It is interesting to note the similarities in chilling effect on the SM and the LM 30 h postmortem pH. It has been shown that the SM and the LM have a large proportion of Type IIb fibers (Christensen et al., 2004; Melody et al., 2004), the current study shows the influ-

Table 3. Effect of chilling on proteolysis and sarcomere length in the *Longissimus thoracic et lumborum*[1]

Item	Conventional chilled sides	Blast chilled sides	SE	*P*-value
Sarcomere length, µm	1.49	1.50	0.01	0.29
Ratio of intact desmin 2 d postmortem[2]	2.27	2.31	0.23	0.87
Ratio of intact desmin 10 d postmortem[2]	1.97	1.75	0.19	0.25
Ratio of degraded troponin-T 2 d postmortem[3]	0.13	0.11	0.01	0.15
Ratio of degraded troponin-T 10 d postmortem[3]	1.02	1.06	0.06	0.52
Calpastatin activity[4]	1.22	1.23	0.05	0.82
Ratio of autolyzed µ-calpain 2 d postmortem[5]	133.10	136.25	4.04	0.44

[1]Least square means reported for each trait.

[2]55 kDa Band Intact Band.

[3]30 kDa Band Degradation Product.

[4]Units of calpastatin activity per gram of tissue 2 d postmortem.

[5]Measured as a ratio of 76 kDa autolysis product of the catalytic subunit.

Table 4. Effect of chilling on desmin degradation in the *Psoas major, Semimembranosus,* and *Triceps brachii*[1]

Ratio of intact desmin	Conventional chilled sides	Blast chilled sides	SE	*P*-value
Psoas major	3.90	3.68	0.19	0.24
2 d postmortem *Psoas major*	3.64	3.74	0.24	0.68
10 d postmortem *Semimembranosus* superficial	2.97	2.95	0.23	0.92
2 d postmortem *Semimembranosus* deep	1.86	2.02	0.19	0.39
2 d postmortem *Semimembranosus* superficial	(0.75)[2] 2.39	(0.63)[2] 2.69	0.30	0.32
10 d postmortem *Semimembranosus* deep	3.46	3.54	0.18	0.67
10 d postmortem *Triceps brachii*	(0.90)[2] 1.68	(0.44)[2] 1.58	0.28	0.71
2 d postmortem *Triceps brachii*	1.85	1.97	0.17	0.49
10 d postmortem				

[1]Least square means reported as ratio of 55 kDa intact desmin band.

[2]*P*-value representation between muscle locations.

Table 5. Effect of chilling on water holding capacity in cuts from aged *Longissimus dorsi, Psoas major, Semimembranosus,* and *Triceps brachii* cuts[1]

Muscle	Conventional chilled sides	Blast chilled sides	SE	*P*-value
Purge loss[2]				
Longissimus dorsi	2.21	2.28	0.33	0.83
Psoas major	0.48	0.74	0.11	0.02
Semimembranosus	3.57	3.52	0.25	0.81
Triceps brachii	1.83	1.82	0.13	0.92
Cook loss[3]				
Longissimus dorsi	22.37	24.24	0.81	0.02
Psoas major	10.99	10.44	0.47	0.24
Semimembranosus	21.92	21.13	0.55	0.16
Triceps brachii	27.06	26.58	0.93	0.60

[1]Least square means of percent loss reported from BC (n = 40) or CC (n = 40) sides.

[2]% Purge Loss measures at time of sensory analysis (LM 10 d, PM 7 d, SM 8 d, TB 13 d) calculated as [(Initial weight – final weight)/initial weight]100.

[3]Cooked to an internal temperature of 68°C; % Calculated as [(raw weight– cooked weight)/raw weight] × 100.

ence of fiber type on postmortem pH may be similar despite chilling method. Differences in pH decline and its relation to fiber type was discussed by Kylä-Puhju et al. (2005). Kylä-Puhju et al. (2005) demonstrated that activity of glucose debranching enzyme (GDE) was lower for darker muscle fibers when compared to lighter muscle fibers. Kylä-Puhju et al. (2005) also showed that low temperature slowed the rate postmortem pH decline, which was potentially confirmed by the present study with LM pH declining at a slower rate in BC sides.

Proteolysis and Sarcomere Length

Sarcomere length can influence water holding capacity and tenderness (Ertbjerg and Puolanne, 2017). In the current study, sarcomere length in the LM was not different across chilling treatments (Table 3) and therefore quality differences cannot be attributed to sarcomere length in the LM. It was hypothesized that rapid chilling would result in less observed protein degradation in the early postmortem period. However, chilling did not affect LM calpain-1 autolysis at 2 d postmortem (Table 3) nor affect intact desmin intensity as measured by the 55 kDa band in the LM (Table 3), PM, SM, or TB measured 2 or 10 d postmortem (Table 4). Furthermore, there was no difference in troponin-T degradation product as measured by the 30 kDa band in the LM aged 2 and 10 d postmortem (Table 3). Calpastatin activity at 2 d postmortem in the LM also was not influenced by chilling treatment (Table 4).

Table 6. Effect of chilling on sensory attributes in aged (10 d) chops from the *Longissimus thoracic et lumborum*[1]

Item	Conventional chilled sides	Blast chilled sides	SE	*P*-value
Marbling score[2]	2.30	2.30	0.15	0.74
Color score[3]	3.10	3.10	0.12	0.92
Hunter L[4]	49.07	49.10	0.53	0.95
Hunter a[4]	13.67	13.79	0.19	0.60
Hunter b[4]	2.90	2.99	0.12	0.43
Star probe, kg[5]	5.45	5.69	0.19	0.21
Warner-Bratzler shear force, kg[6]	2.00	2.30	0.15	0.05
Juiciness[7]	7.00	7.00	0.21	0.75
Tenderness[7]	7.00	6.60	0.28	0.20
Chewiness[7]	3.70	4.00	0.34	0.35
Flavor[7]	3.60	3.60	0.15	0.94
Off-flavor[7]	1.50	1.30	0.17	0.18

[1]Least square means reported for each trait.

[2]National Pork Board standards, 10-point scale (1 = 1.0% intramuscular fat; 10 = 10% intramuscular fat).

[3]National Pork Board standards, 6-point scale (1 = pale pinkish gray to white; 6 = dark purplish red).

[4]Hunter L a b, D65 light source, 50 mm aperture, 2° observer.

[5]Force required to compress sample to 20% of its original height.

[6]Force required to shear 1.27 cm core.

[7]Trained sensory analysis (n = 4) samples scored on a 10-point scale. Lower scores indicate less juiciness, tenderness, chewiness, flavor, and off-flavor.

Table 7. Effect of chilling on sensory attributes in aged (7 d) roasts from the *Psoas major*[1]

Item	Conventional chilled sides	Blast chilled sides	SE	*P*-value
Color score[2]	3.80	4.00	0.13	0.13
Hunter L[3]	40.35	40.34	0.68	0.98
Hunter a[3]	14.68	14.59	0.32	0.79
Hunter b[3]	1.35	1.36	0.15	0.93
Star probe, kg[4]	2.40	2.39	0.09	0.91
Warner-Bratzler shear force, kg[5]	2.54	2.47	0.10	0.48
Juiciness[6]	7.50	8.30	0.17	< 0.0001
Tenderness[6]	7.90	8.60	0.25	0.005
Chewiness[6]	2.80	2.10	0.20	0.002
Flavor[6]	3.50	3.70	0.13	0.13
Off-flavor[6]	1.90	1.90	0.15	0.93

[1]Least square means reported for each trait.

[2]National Pork Board standards, 6-point scale (1 = pale pinkish gray to white; 6 = dark purplish red).

[3]Hunter L a b, D65 light source, 50 mm aperture, 2° observer.

[4]Force required to compress sample to 20% of its original height.

[5]Force required to shear 1.27 cm core.

[6]Trained sensory analysis ($n = 4$) samples scored on a 10 point scale. Lower scores indicate less juiciness, tenderness, chewiness, flavor, and off-flavor.

Table 8. Effect of chilling on sensory attributes in aged (8 d) chops from the *Semimembranosus*[1]

Item	Conventional chilled sides	Blast chilled sides	SE	*P*-value
Marbling score[2]	2.30	2.40	0.12	0.31
Color score[3]	3.00	3.20	0.09	0.05
Hunter L[4]	41.76	41.20	0.64	0.39
Hunter a[4]	16.35	16.02	0.16	0.05
Hunter b[4]	3.11	2.93	0.13	0.17
Superficial star probe, kg[5]	5.79	5.71	0.17	0.63
Deep star probe, kg[5]	5.83 (0.81)[6]	5.88 (0.35)[6]	0.16	0.78
Superficial Warner-Bratzler shear force, kg[7]	4.28	4.07	0.29	0.50
Deep Warner-Bratzler shear force[7]	4.21 (0.80)[6]	4.38 (0.29)[6]	0.20	0.53
Juiciness[8]	6.90	6.90	0.10	0.80
Tenderness[8]	6.10	5.90	0.20	0.38
Chewiness[8]	4.80	5.10	0.06	0.34
Flavor[8]	3.40	3.50	0.14	0.38
Off-flavor[8]	1.40	1.30	0.28	0.07

[1]Least square means reported for each trait.

[2]National Pork Board standards, 10-point scale (1 = 1.0% intramuscular fat; 10 = 10% intramuscular fat).

[3]National Pork Board standards, 6-point scale (1 = pale pinkish gray to white; 6 = dark purplish red).

[4]Hunter L a b, D65 light source, 50 mm aperture, 2° observer.

[5]Force required to compress sample to 20% of its original height.

[6]*P*-value representative between muscle locations. Fixed effects of TRT, harvest, harvest*TRT, muscle, and muscle*TRT with carcass serving as random effect used for analysis. Least square means for this model represented in table.

[7]Force required to shear 1.27 cm core.

[8]Trained sensory analysis ($n = 4$) samples scored on a 10-point scale. Lower scores indicate less juiciness, tenderness, chewiness, flavor, and off-flavor.

Water holding capacity

Least squares means of purge loss and cook loss are summarized in Table 5. Chilling regime did not have an effect on purge loss in cuts from the LM, SM or TB. These results are in contrast to other reports that reported rapid chilling resulted in less purge and drip loss (McFarlane and Unruh, 1996; Janiszewsi et al., 2018). The difference may be that the control methods in those investigations had a slower chilling rate.

Roasts from the PM of BC sides had greater drip loss than CC sides. Because of the PM's exposed location in a dressed carcass, it is less insulated and it is likely that temperature decline will be most rapid in this muscle, and will experience a cold shortening effect that will decrease myofibrillar space for water storage (Honikel et al., 1986; Offer and Cousins, 1992). Offer and Cousins (1992) found that the formation of drip in the event of sarcomere shortening could be explained by the formation of "drip channels" allowing water to escape from muscle fiber to the endomysium and ultimately lost from muscle tissue. This could explain the increased purge loss in roasts from the PM of BC sides. Although sarcomere length was not measured in these samples, a discussion point is that more rapid chilling in the PM might have created a cold shortening effect, creating drip channels and increasing purge lost.

Increased purge in the PM could also be explained by formation of ice crystals on the surface of muscle tissue. Rapid chilling of pork, specifically by air chilling, has been found to lead to ice crystal formation throughout muscle structure (Ngapo et al., 1999; Hansen et al., 2003). When muscle thaws, there is a significant increase in purge loss due to ice formation and melting.

Linkage of myofibrils in the LM may have an effect on water loss during cooking due to sarcomere shortening. This greater contraction during chilling could be a result of lack of early postmortem degradation of intermediate filament proteins which link muscle fibers to the sarcolemma and other muscle fibers at the Z-line causing increased purge loss (Morrison et al., 1998; Kristensen and Purslow, 2001; Melody et al., 2004; Bee et al., 2007). Melody et al. (2004) found that degradation of desmin in the PM began as early as 45 min postmortem in conventionally chilled pork carcass. Pomponio et al. (2010) found that early postmortem pH decline influenced μ-calpain activity with muscle with faster postmortem

Table 9. Effect of chilling on sensory attributes in aged (13 d) roasts from the *Triceps brachii*[1]

Item	Conventional chilled sides	Blast chilled sides	SE	*P*-value
Color score[2]	2.90	2.90	0.10	0.62
Hunter L[3]	41.79	41.70	0.42	0.82
Hunter a[3]	16.03	16.02	0.13	0.95
Hunter b[3]	4.35	4.35	0.12	0.99
Star probe, kg[4]	4.93	5.00	0.15	0.64
Warner-Bratzler shear force, kg[5]	2.91	3.10	0.13	0.16
Juiciness[6]	5.40	5.70	0.22	0.25
Tenderness[6]	4.70	4.70	0.75	0.99
Chewiness[6]	5.50	5.80	0.26	0.27
Flavor[6]	4.00	3.90	0.10	0.18
Off-flavor[6]	1.30	1.20	0.05	0.32

[1]Least square means reported for each trait.

[2]National Pork Board standards, 6-point scale (1 = pale pinkish gray to white; 6 = dark purplish red).

[3]Hunter L a b, D65 light source, 50 mm aperture, 2° observer.

[4]Force required to compress sample to 20% of its original height.

[5]Force required to shear 1.27 cm core.

[6]Trained sensory analysis ($n = 4$) samples scored on a 10-point scale. Lower scores indicate less juiciness, tenderness, chewiness, flavor, and off-flavor.

pH decline having increased μ-calpain activity postmortem. No difference in desmin degradation was noted at 2 or 10 d in due to treatment in the current study, but no commentary can be provided for the contribution to rate of proteolysis before the 2 d postmortem.

Sensory and Texture analysis

Chops from the LM of BC sides had greater cook loss (Table 5) and WBS (Table 6) than chops from CC sides but demonstrated no significant difference in proteolysis, calpastatin activity or calpain autolysis (Table 3). Results from the current study are consistent with previous results (Shackelford et al., 2012) that also found an increased toughness in the LM as determined by slice shear force value but found no difference in proteolysis after 15 d aging. Increased WBS values were not reflected in star probe values, or for sensory tenderness or chewiness.

Roasts from the PM of BC sides were juicier, more tender and less chewy than roasts from CC sides (Table 7) but treatment did not affect WBS or star probe. Blast chilling had an effect in the SM with decreased Hunter a value indicating a lower intensity of red pigment, and a darker observed color score (Table 8). No treatment effect was found in cuts from the TB (Table 9).

Postmortem pH decline, protein degradation, and changing postmortem muscle structure impacts fresh pork quality. The current study confirms the influence of chilling on fresh pork quality varies with muscle metabolism and carcass location. Postmortem proteolysis and changes in muscle biochemistry are responsible for the development of fresh pork quality. Slowing the rate of postmortem pH decline in the LM has a direct effect on the rate and extent of normal changes in the conversion of muscle to meat. These changes include protein degradation and denaturation. Rapid chilling did result in greater WBS in LM chops, but did not affect star probe, sensory tenderness, or sarcomere length. In addition, no difference in proteolysis due to chilling protocol was detected in any muscle evaluated. The influence of chilling on early postmortem muscle modification may warrant further investigation.

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